

## Boron Balance in Humans

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Studies of boron (B) in animals and humans indicate that boron homeostasis is maintained by urinary excretion. A relationship between dietary boron and bone mineral metabolism also has been reported. To understand further boron metabolism, boron balance and urinary calcium excretion were measured in seven healthy men who were participating in a controlled, metabolic study of zinc homeostasis. The 20-week study consisted of four metabolic periods: equilibration, 3 weeks; baseline, 2 weeks; low zinc period, 10 weeks; zinc repletion, 5 weeks. A diet of conventional foods provided a daily average boron intake of 3.73 mg B. Dietary zinc intake was 13.7 mg/d during the equilibration, baseline and repletion periods, and 4.6 mg/d during the low zinc period. Urinary excretion of boron and calcium was measured for 8 weeks during the study—the second week of the baseline period, the first 2 weeks of the low zinc period, and the entire 5 weeks of the zinc repletion period. Fecal boron excretion and boron balance [dietary boron less (urinary boron + fecal boron)] were measured for 6 weeks during the study—the second week of the baseline period, the first 2 weeks of the low zinc period, and the last 3 weeks of the 5-week zinc repletion period. Daily urinary and fecal boron excretion averaged 3.18 mg B (85% of dietary boron) and 0.29 mg B (8% of dietary boron), respectively. The subjects were in slight positive boron balance (0.19 mg B/d). Urinary excretion of calcium during the 5-week repletion period was significantly ( $P < 0.05$ ) higher, on average 42%, than during the 3 weeks at the end of baseline and at the beginning of the low zinc period. Urinary boron excretion was significantly ( $P < 0.05$ ) higher during weeks 3 and 4 of the repletion period than during the first 2 weeks. Boron homeostasis appears to be regulated by the kidney, and urinary boron excretion responds quickly to changes in dietary boron. Changes in dietary zinc did not alter urinary boron excretion. There is no evidence that the elevated urinary calcium excretion, seen during the entire zinc repletion period, is related to boron metabolism. *J. Trace Elem. Med.* 12:271–284, 1999. © 1999 Wiley-Liss, Inc.

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## INTRODUCTION

In humans, homeostatic mechanisms respond to changes in dietary intakes of nutrients by regulating gastrointestinal absorption and/or urinary losses. Animal and human studies of boron excretion suggest that boron homeostasis is maintained by urinary excretion. Farr and Konikowski [1] reported that mice dosed intravenously with either 0.5 mg or 2.1 mg of boron, as sodium pentaborate, excreted about half the administered dose in the urine within 1 hour. Vanderpool et al. [2] measured the rate of boron excretion using the stable isotope of boron,  $^{10}\text{B}$ . They demonstrated that 95% of a 20- $\mu\text{g}$  dose of  $^{10}\text{B}$ , fed to rats in a test meal, could be detected in the urine within 3 days. Only 4% of the  $^{10}\text{B}$  dose appeared in the feces. A similar excretory pattern has been shown in humans. Schou et al. [3] reported that men given 750 mg of boric acid dissolved in water excreted 94% of the dose in the urine over 4 days. It is important to note, however, that these studies all employed the administration of boric acid or borates as pure chemicals either alone or added to diets and thus not as constituents of customary foods and beverages.

Boron balance studies in humans also show that boron is well absorbed and rapidly excreted in the urine. The first human boron balance study, reported in 1941 by Kent and McCance [4], was done in two women who were participating in a metabolic study of mineral availability from diets in which bread provided 4% of the calories. Boron balance was calculated over 4 consecutive weeks. When the women were given a 352 mg dose of boron, as boric acid, in addition to the diet, 93% of the boron intake was excreted in the urine during the first week, whereas very little appeared in the feces. When the women's boron intake was from diet alone, the urinary boron excretion was 83–99% of intake. The results of this study suggested that boron in foods is metabolized in the same way as a single boron dose.

In 1997, Hunt et al. [5] reported the boron losses in urine and feces of 11 postmenopausal women who were participating in a 24-week metabolic study. Two dietary levels of boron were fed during the study: a low level of  $<0.36$  mg boron/d for 17 weeks and a supplemented level of 3.23 mg boron/d for 7 weeks. When the dietary boron was  $<0.36$  mg/d, the mean urinary and fecal boron excretion averaged  $0.37 \pm 0.15$  mg/d and  $0.05 \pm 0.19$  mg/d, respectively. The subjects were in a slight negative boron balance; an average of  $-0.06$  mg boron was lost each day. When boron intake was increased nine fold to 3.23 mg/d, the mean urinary and fecal excretion of boron was  $2.87 \pm 0.15$  mg/d and  $0.11 \pm 0.06$  mg/d, respectively. The subjects were in a slight positive balance of 0.25 mg boron/d. The findings of these two studies show that in humans, boron is well absorbed and excreted primarily by the kidneys. Hunt et al. [5] also reported that a boron intake of 3.23 mg/d was associated with a decrease in the percentage of dietary calcium excreted in the urine of the postmenopausal women, suggesting an interaction between boron and bone mineral metabolism.

To investigate further the relationship among dietary boron, boron homeostasis, and the interaction of boron with calcium metabolism, we measured urinary and fecal boron losses and urinary calcium excretion in seven Caucasian men who were participating in a controlled, metabolic study of zinc homeostasis. The objectives of the study were: to (1) measure the boron balance in men fed a constant boron intake for 17 weeks, (2) determine if changes in dietary zinc alter boron excretion, (3) compare the variability of urinary boron excretion with the urinary excretion of sodium and

creatinine, and (4) investigate the relationship of boron with calcium and bone metabolism.

## MATERIALS AND METHODS

The study was performed at the Metabolic Research Unit (MRU) of the U.S. Department of Agriculture (Western Human Nutrition Research Center, San Francisco, CA). The 20-week study consisted of four metabolic periods: a 3-week equilibration period, a 2-week baseline period, a 10-week low zinc period, and a 5-week zinc repletion period (Table 1). During the equilibration period, the subjects lived at home and went daily to the MRU for lunch and dinner, and they took home their evening snack and breakfast for the following morning. After completing the equilibration period, the subjects were confined to the MRU for 17 weeks (baseline, low zinc, and zinc repletion periods). During confinement, the men walked 3 miles at a moderate pace, twice daily, to maintain good muscle tone, physical fitness, and energy balance. In addition, an individualized daily exercise schedule was planned for each subject to maintain his usual activity pattern, and the subjects were escorted whenever they left the MRU. The study was approved by the Human Subjects Institutional Review Boards of the University of California, Berkeley, and the U.S. Department of Agriculture. Written informed consent was obtained from each subject.

### Study Diet

The study diet, which began at the equilibration period, consisted of conventional foods and beverages. Three different menu days were repeated throughout the study (Table II). Components of the diet are shown in Table III. The study diet provided, on average, 3.73 mg of boron and 4.6 mg of zinc per day. During the equilibration, baseline, and repletion periods, the subjects received a zinc supplement (9.03 mg zinc/d, as zinc gluconate, prepared by Drug Product Service Lab, University of California, San Francisco), which brought the total zinc intake to 13.7 mg/day. The zinc gluconate was added to capsules containing para-aminobenzoic acid (PABA) (Drug Product Service Lab), which was given throughout the study to monitor the completeness of urine collections. Each subject received three capsules/day, each containing 80 mg PABA and 3.01 mg zinc per capsule. During the low zinc period, capsules containing only PABA were given. The PABA and PABA + zinc capsules were identical in appearance, and the subjects were unaware of the change in metabolic periods. Boron intake for menu day 1, 2, and 3 was  $4.56 \pm 0.1$  mg/d,  $1.87 \pm 0.1$  mg/d, and  $4.75 \pm 0.1$  mg/d, respectively. The higher boron content of menu days 1 and

TABLE I. Study Design

Metabolic period	Duration (weeks)	Collection times		Dietary zinc (mg/d)	Dietary boron (mg/d)
		Urine (weeks of metabolic period)	Feces		
Equilibration	3	—	—	13.7	>3.73
Baseline	2	2	2	13.7	3.73
Low zinc	10	1–2	1–2	4.6	3.73
Zinc repletion	5	1–5	3–5	13.7	3.73

**TABLE II. Three-Day Cycle Menu**

Menu day 1	Menu day 2	Menu day 3
<u>Breakfast</u> cornflakes pineapple juice* fruit cocktail* milk 2%	<u>Breakfast</u> corn chex orange juice* blueberries milk 2%	<u>Breakfast</u> frosted flakes wheaties apple juice* peaches* milk 2%
<u>Lunch</u> croissant mayonnaise ham lettuce safflower oil vinegar dill pickle pears, canned*	<u>Lunch</u> chicken breast pasta Italian dressing lettuce safflower oil vinegar pound cake strawberries, frozen cool whip sugar	<u>Lunch</u> chicken breast white rice safflower oil broccoli lettuce safflower oil vinegar bread roll margarine sugar cookie
<u>Dinner</u> chicken breast chicken gravy safflower oil pasta green beans pudding mix, choc cool whip	<u>Dinner</u> chicken breast cr. chicken soup, cond safflower oil potato carrots biscuit margarine pineapple, canned*	<u>Dinner</u> chicken breast potatoes, flakes water margarine green beans gravy pears, canned*
<u>Evening snack</u> vanilla wafer grape juice*	<u>Evening snack</u> shortbread cookie fruit punch	<u>Evening snack</u> oatmeal cookie lemonade
<u>Boron intake (mg)</u> 4.56 ± 0.1	<u>Boron intake (mg)</u> 1.87 ± 0.1	<u>Boron intake (mg)</u> 4.75 ± 0.1

\*Foods of high boron content.

**TABLE III. Diet Components\***

Basic menu, 3 day cycle
Extra energy formula (oil, sugar, dextro-maltose)
Protein supplement
Iron supplement, Fer-In-Sol
Calcium supplement, Tums
Magnesium supplement, MgO
PABA <sup>a</sup>
PABA + Zn gluconate <sup>b</sup>

\*Prepared by Drug Product Service Lab, University of California, San Francisco.

<sup>a</sup>PABA (Para-aminobenzoic acid), low Zn period only.<sup>b</sup>PABA + Zn gluconate, baseline, and repletion.

3 was due to the inclusion of boron-rich fruits and juice (pears, peaches, apple juice, pineapple juice, and grape juice).

Individual energy needs were met by the addition of an extra-energy formula: oil (Saffola), sugar, dextro-maltose (Saroni, Oakland, CA). During the baseline period, the energy intake was adjusted to maintain constant body weight. Thereafter, the energy level of the diet for each subject was kept constant for the remainder of the study. Energy intakes ranged from 2,917–3,493 kcal/d (39–57 kcal/kg body wt/d). Egg albumen powder (Saroni) was added to the diet to provide at least 0.8 g protein/kg body weight for each subject; protein intakes ranged from 62–74 g/d (0.8–1.1 g/kg body wt/d). Magnesium, calcium, and iron supplements were added to the diet to meet the RDAs for those minerals; the intakes of all other nutrients either met or exceeded the RDAs from the diet alone. A database nutrient analysis (USDA, Agriculture Handbook No. 8, Version 10, Series 1–21, 1991) of the diet is shown in Table IV. Deionized water was available for drinking as desired, and the amount consumed by each subject was recorded daily.

### Sample Collections and Analysis

**Diet.** Composites from each menu day were prepared four times during the study: once during the equilibration (days 12–16) and repletion (days 27–31) periods and twice during the repletion period (days 20–25 and 62–66) (Table V). Aliquots from each composite and the additional dietary items (oil, sugar, egg albumen, and mineral supplements) were analyzed for boron and zinc. The composites were stored at  $-20^{\circ}\text{C}$  until analyzed.

**Blood.** Fasting venous blood samples were drawn from the antecubital vein of the recumbent subjects for weekly measurement of plasma zinc concentration and other blood parameters—complete blood count (CBC) and sequential multiple analysis (SMAC) blood panels—to monitor zinc and health status. Fasting blood samples for zinc analysis were drawn into Sarstedt (Newton, NC) syringes containing ammonium heparin-coated beads (15 I.U. heparin/mL blood). Blood samples for the measurement of blood components other than zinc were drawn into trace element-free Vacutainers (Sarstedt) containing the appropriate preservative. All blood samples were placed on ice for no longer than 60 minutes and then centrifuged in a Sorvall RC-5C centrifuge (Sorvall® Instruments, Dupont Corp., Wilmington, DE) under refrigeration for 10 minutes at 2,400 rpm (1,145 $\times$ g). Samples were stored at  $-70^{\circ}\text{C}$  until analyzed.

**Urine and feces.** Twenty-four-hour urine and fecal collections were made throughout the study. Each 24-hour collection began at 0800 hours on the first morning and finished at 0800 hours the following morning. Urine aliquots for analysis of boron and calcium were made during the second week of baseline, the first 2 weeks of the low zinc period, and the entire 5 weeks of the zinc repletion period. Urine aliquots for zinc analysis were made from each daily collection. All samples were stored at  $-20^{\circ}\text{C}$  until analyzed. For fecal boron analysis, aliquots of the daily fecal collections were combined into weekly fecal pools during the second week of baseline, the first 2 weeks of the low zinc period, and the last 3 weeks of the zinc repletion period.

**Boron analysis.** Boron analysis was performed by West Coast Analytical Services (Santa Fe Springs, CA). Urine samples were diluted 10-fold with nitric acid. Aliquots from each diet composite and seven-day fecal pool were lyophilized and homogenized, then digested in sealed Teflon vessels using a microwave digestion system.

**TABLE IV. Average Nutrient Composition of Diet From Nutrient Database and Supplements**

Nutrient	Baseline	Metabolic period	
		Low zinc	Zinc repletion
Energy (Kcals)	3,310.54	3,264.26	3,262.21
Protein (g)	65.39	65.42	65.41
Fat (g)	117.25	116.78	116.50
Carbohydrate (g)	517.13	505.49	505.55
Fiber (g)	21.59	21.12	21.20
Calcium (mg)	611.84	613.05	614.10
Iron (mg)	17.01	17.03	17.02
Magnesium (mg)	267.30	266.33	266.49
Phosphorus (mg)	955.25	955.19	955.30
Zinc, analysis (mg)	13.67	4.63	13.66
Copper (mg)	1.47	1.47	1.47
Manganese (mg)	5.17	5.21	5.21
Potassium (mg)	3,087.97	3,076.21	3,078.49
Sodium (mg)	2,771.39	2,842.82	2,835.66
Pantothenic acid (mg)	4.22	4.23	4.23
Ascorbic acid (mg)	213.61	233.13	229.87
Thiamin (mg)	2.46	2.49	2.48
Riboflavin (mg)	2.23	2.19	2.20
Niacin (mg)	32.24	32.37	32.35
Pyridoxine (mg)	2.68	2.69	2.68
Folacin ( $\mu$ g)	440.43	445.36	444.54
Vitamin B12 ( $\mu$ g)	2.91	3.11	3.07
Vitamin A (RE)	1,555.26	1,568.61	1,566.39
Vitamin E (mg a-te)	23.59	23.04	23.01
Saturated fat (g)	27.54	27.41	27.41
Mono-unsaturated fat (g)	54.50	53.93	53.76
Poly-unsaturated fat (g)	27.69	27.91	27.83
Cholesterol (mg)	130.89	130.37	130.45
Supplements	Product	Nutrient	
Iron (FeSO <sub>4</sub> ) <sup>a</sup>	Fer-In-Sol	3 mg Fe/d	
Magnesium (MgO) <sup>b</sup>	Magnesium	125 mg Mg/d	
Calcium (CaCO <sub>3</sub> ) <sup>c</sup>	Tums	200 mg Ca/d	

<sup>a</sup>Fer-In-Sol, Mead Johnson, Evansville, IN.<sup>b</sup>Magnesium, Longs Drug Stores, Walnut Creek, CA.<sup>c</sup>Tums, SmithKline Beecham, Pittsburgh, PA.**TABLE V. Subject Characteristics**

Subject	Age (y)	Weight (kg)	Height (cm)	BMI (kg/m <sup>2</sup> )	Ethnicity
2	34	90.1	169.2	31	Caucasian
3	42	68.0	168.5	24	Caucasian
4	27	74.6	186.2	22	Caucasian
7	37	79.3	177.2	25	Caucasian
8	47	57.1	167.6	20	Caucasian
9	44	78.5	187.0	22	Caucasian
10	28	71.1	182.2	21	Caucasian
Mean	37	74.1	176.8	24	
S.D.	8	10.3	8.5	4	

The digest was brought up to volume with nitric acid. All samples were analyzed in duplicate by inductively coupled plasma-mass spectrometry (ICP-MS).

**Zinc and calcium analysis.** Plasma and urinary zinc and urinary calcium analyses were performed on duplicate samples by atomic absorption spectrophotometry using a Smith-Hieftje-22 Atomic Absorption Spectrophotometer (Thermo Jarrell Ash Corp., Franklin, MA) with background correction. Reliability of the method was verified by analysis of a bovine liver sample obtained from the National Institute of Standards and Technology (U.S. Department of Commerce, National Institute of Standards and Technology, Gaithersburg, MD).

**Sodium.** Urinary sodium analysis was performed using a Nova 5 Electrolyte Analyzer (Nova Biomedical, Waltham, MA) with ion-specific electrodes to measure sodium.

**Creatinine.** Daily urinary creatinine was measured with an alkaline picrate kinetic method using an automated centrifugal analyzer (Cobas-Fara 2, Roche Diagnostic Systems, Nutley, NY).

**Boron balance.** Boron balance was calculated by subtracting the daily urinary and fecal boron losses from the daily boron intake. The balance calculation did not include integumental and miscellaneous boron losses. These losses were not estimated because values for boron losses in sweat, sloughed skin, and other components of integumental and miscellaneous losses have not been measured or reported.

### Statistical Analysis

Comparisons between metabolic periods were made by repeated measures analysis of variance. If statistical differences were found, the Tukey's studentized range t-test was performed to determine which time points differed from each other. Analysis of urinary calcium values was performed on logarithm transformed data. Within subject, relationships were determined using repeated measure analysis of covariance with covariance changing over time. Analysis were considered statistically significant with a *P* value <0.05.

## RESULTS

### Subjects

Seven healthy men participated in the low zinc homeostasis study. Their ages ranged from 27–47 years, they weighed an average of  $75.4 \pm 10.2$  kg, were  $177 \pm 8$  cm tall, and had a body mass index (BMI) that ranged from 20–31 kg/m<sup>2</sup> (Table V). All of the men were Caucasian and had no recent history of cigarette, drug, or alcohol abuse, no recent use of mineral supplements, and no history of medical problems that would alter Zn status (trauma, infection, diabetes mellitus, hormonal disorders). They all reported having a stable body weight, and their usual diet included red meat several times a week. All subjects were considered to be in good health based on medical histories and physical examinations performed by a physician. All the subjects remained in good health during the study. Over the study period, the subjects' body weight increased gradually, probably due to a decrease in physical activity as a result of confinement. Total body weight at the end of zinc repletion ( $77.9 \pm 10.7$  kg) was significantly higher (*P* < 0.05) than the baseline value,  $75.7 \pm 10.6$  kg.



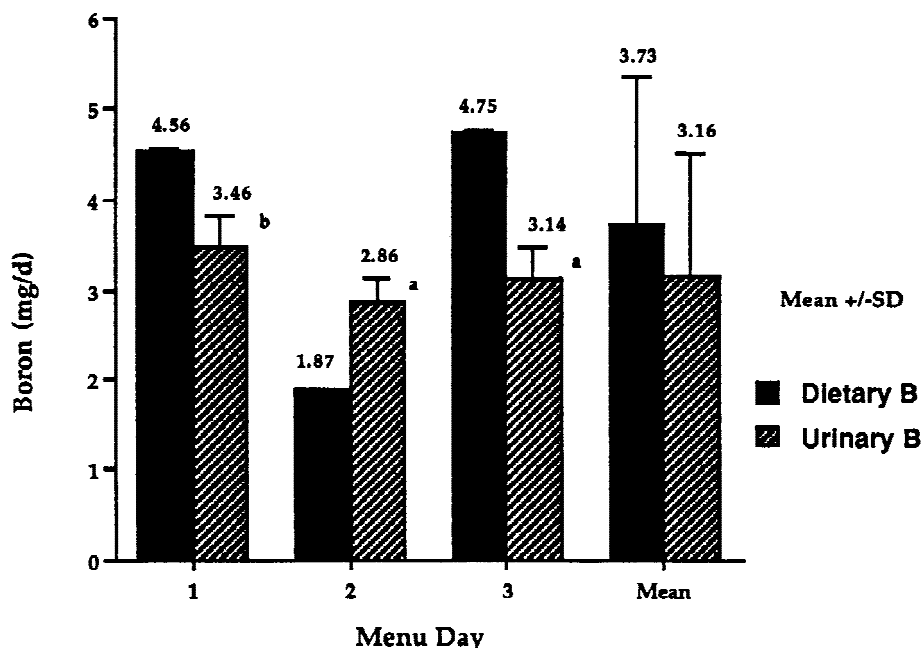
### Zinc Status

Plasma zinc concentration did not change significantly throughout the study. Mean plasma zinc concentration at the end of the baseline, low zinc, and zinc repletion periods was  $73.4 \pm 8.4$   $\mu\text{g/dL}$ ,  $73.0 \pm 9.2$   $\mu\text{g/dL}$ , and  $78.1 \pm 7.1$   $\mu\text{g/dL}$ , respectively. The 6-day average urinary zinc excretion at the end of the baseline, low zinc, and zinc repletion periods was  $0.42 \pm 0.21$  mg/d,  $0.42 \pm 0.21$  mg/d and  $0.48 \pm 0.22$  mg/d, respectively. There was no significant change in urinary zinc excretion throughout the study.

### Boron Excretion and Balance

**Boron excretion.** Urinary boron excretion responded readily to changes in dietary boron over the 3 menu days. Urinary boron excretion for menu days 1, 2, and 3 averaged  $3.46 \pm 0.35$ ,  $2.86 \pm 0.28$ , and  $3.14 \pm 0.32$  mg/d, respectively (Fig. 1). Dietary boron was high on menu day 1 (4.56 mg/d) and menu day 3 (4.75 mg/d). The urinary boron excretion on these days was significantly higher than on menu day two ( $P < 0.05$ ). To account for the variation in urinary boron excretion associated with the variation in dietary boron intake, two menu cycles of 3 days each (i.e., 6 days) were averaged to compute urinary boron excretion.

The average daily urinary and fecal boron excretion over each collection period during the study is shown in Figure 2. Urinary boron excretion during the 8 weeks in which samples were collected averaged 3.18 mg/d, representing 85% of average dietary boron intake. Urinary boron excretion did not change with decreases or in-



**Fig. 1.** Dietary boron intake and urinary boron excretion. (a) and (b) indicate significant statistical difference at  $P < 0.05$ .



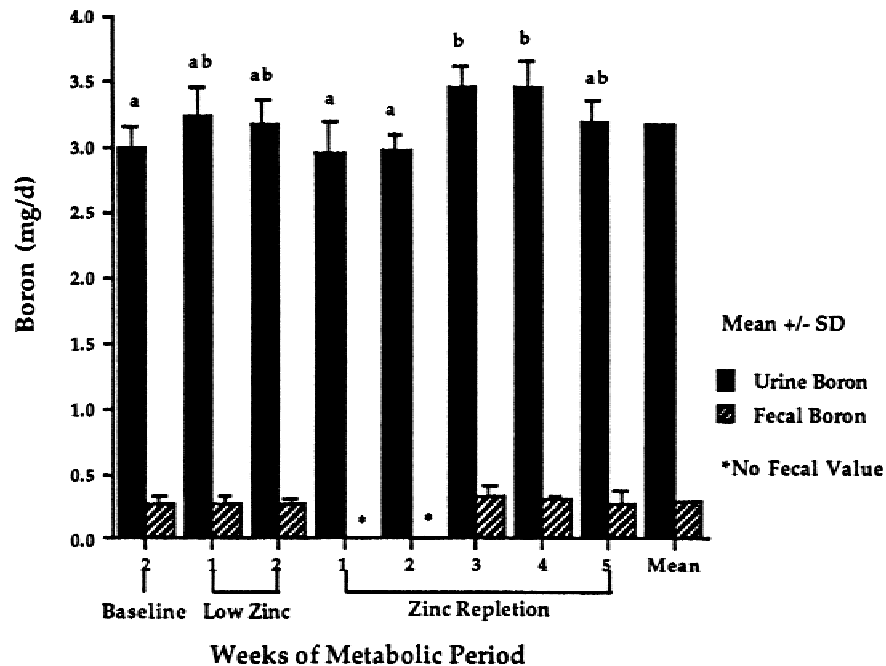


Fig. 2. Urinary and fecal boron excretion. (a), (b), and (ab) indicate significant statistical difference at  $P < 0.05$ . No fecal values at repletion weeks 1 and 2 because samples were not collected at these times.

creases in dietary zinc. However, urinary boron excretion was significantly higher ( $P < 0.05$ ) during the third and fourth week of zinc repletion than during baseline and the first 2 weeks of zinc repletion (Table VI). Fecal boron excretion during the 6 weeks of collection averaged 0.29 mg/d, which represents 8% of average dietary boron intake. Fecal boron excretion did not change with changes in dietary zinc, but it also tended to increase during the third and fourth week of zinc repletion; this change was not significant, however (see Fig. 4).

Individual urinary and fecal boron excretion, averaged over 6 days at each time point, is presented in Figures 3 and 4. Urinary boron excretion of all subjects over the entire set of collections ranged from 2.58 mg B/d–3.68 mg B/d, or 69–99% of boron intake. The within-subject coefficient of variation of urinary boron excretion ranged from 16–25%. This variation in urinary boron excretion was not related to water intake, urine volume, or urinary sodium, calcium, zinc, or creatinine excretion (Table VI). The within-subject coefficient of variation, over the corresponding period, for daily urinary excretion of sodium ranged from 17–24% and for creatinine ranged from 7–18%.

Individual fecal boron losses over the entire set of collections ranged from 0.14–0.47 mg/d, or 4–13% of dietary boron intake. The individual variability in fecal boron excretion was 8–32%, which probably reflects individual differences in gastrointestinal transit time.

**Boron balance.** Boron balance, calculated as the difference between boron intake and the boron losses in the urine and feces, ranged from 0.45 mg/d–0.05 mg/d during the 6 weeks that samples were collected (Fig. 5). Inclusion of boron losses in integu-

**TABLE VI. Daily Water Intake, Urine Volume, and Urinary Excretion of Boron, Sodium, Calcium, Zinc, and Creatinine (mean  $\pm$  S.D.)**

Metabolic period week	Baseline 2	Low zinc		Zinc repletion				
		1	2	1	2	3	4	5
Water intake (L/d) <sup>1</sup>	1.29 $\pm$ 0.44	1.35 $\pm$ 0.47	1.73 $\pm$ 0.39	1.39 $\pm$ 0.35	1.32 $\pm$ 0.40	1.33 $\pm$ 0.47	1.24 $\pm$ 0.35	1.37 $\pm$ 0.62
Urine volume (L/d)	2.31 $\pm$ 0.65	2.67 $\pm$ 0.63	2.92 $\pm$ 0.54	2.44 $\pm$ 0.59	2.40 $\pm$ 0.57	2.53 $\pm$ 0.67	2.56 $\pm$ 0.58	2.69 $\pm$ 0.70
Boron (mg/d) <sup>2</sup>	3.01 $\pm$ 0.16 <sup>a</sup>	3.25 $\pm$ 0.22 <sup>abc</sup>	3.18 $\pm$ 0.17 <sup>ab</sup>	2.95 $\pm$ 0.25 <sup>a</sup>	2.97 $\pm$ 0.13 <sup>a</sup>	3.46 $\pm$ 0.17 <sup>c</sup>	3.45 $\pm$ 0.22 <sup>bc</sup>	3.19 $\pm$ 0.17 <sup>ab</sup>
Sodium (g/d)	1.88 $\pm$ 0.26	2.22 $\pm$ 0.11	2.20 $\pm$ 0.25	1.97 $\pm$ 0.27	2.03 $\pm$ 0.32	2.04 $\pm$ 0.42	2.19 $\pm$ 0.56	1.97 $\pm$ 0.48
Caclium (mg/d) <sup>2*</sup>	144.3 $\pm$ 88.0 <sup>a</sup>	154.4 $\pm$ 86.9 <sup>ab</sup>	142.7 $\pm$ 71.2 <sup>a</sup>	201.4 $\pm$ 99.5 <sup>c</sup>	201.0 $\pm$ 89.3 <sup>c</sup>	204.3 $\pm$ 90.4 <sup>c</sup>	198.3 $\pm$ 101.0 <sup>bc</sup>	216.1 $\pm$ 102.8 <sup>c</sup>
Zinc (mg/d)	0.42 $\pm$ 0.21	0.37 $\pm$ 0.16	0.40 $\pm$ 0.18	0.41 $\pm$ 0.21	0.40 $\pm$ 0.22	0.46 $\pm$ 0.24	0.45 $\pm$ 0.19	0.48 $\pm$ 0.22
Creatinine (g/d)	1.54 $\pm$ 0.24	1.56 $\pm$ 0.21	1.55 $\pm$ 0.20	1.50 $\pm$ 0.21	1.51 $\pm$ 0.20	1.55 $\pm$ 0.22	1.53 $\pm$ 0.25	1.57 $\pm$ 0.25

<sup>1</sup>Does not include liquids with diet.<sup>2</sup>n = 5.\*Values not sharing the same superscript are significantly different at  $P < 0.05$ .

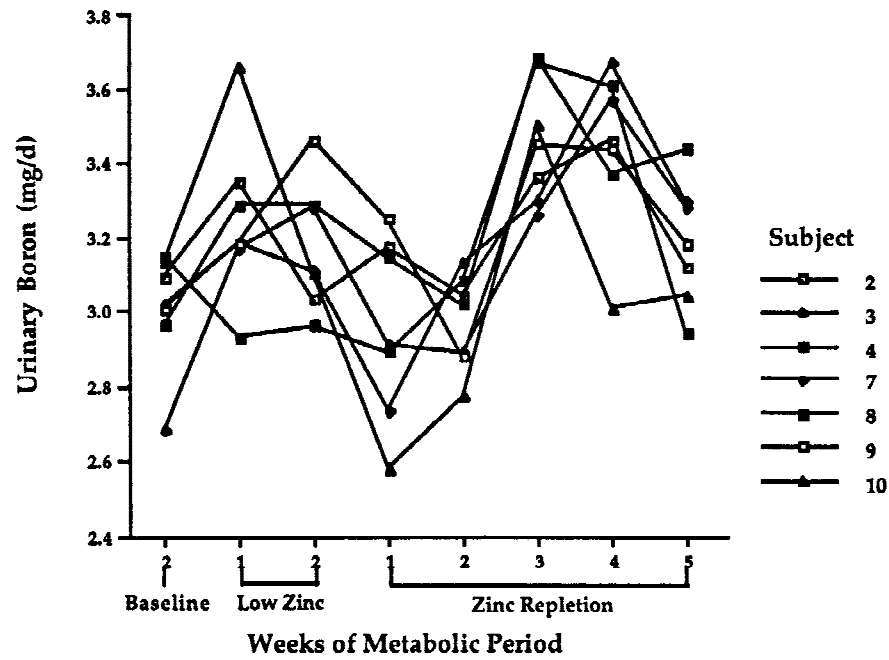


Fig. 3. Subject's urinary boron excretion.

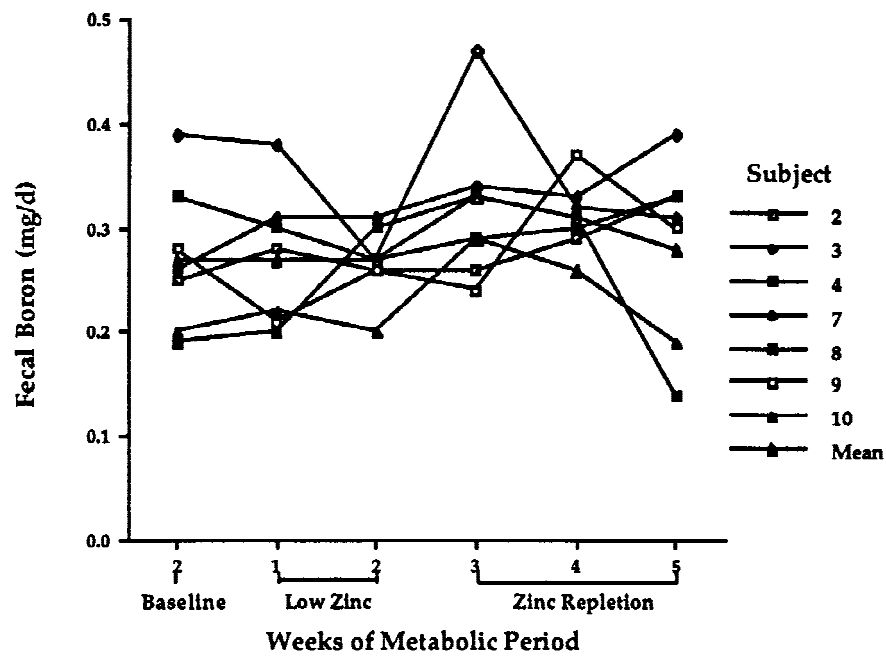


Fig. 4. Subject's fecal boron excretion.

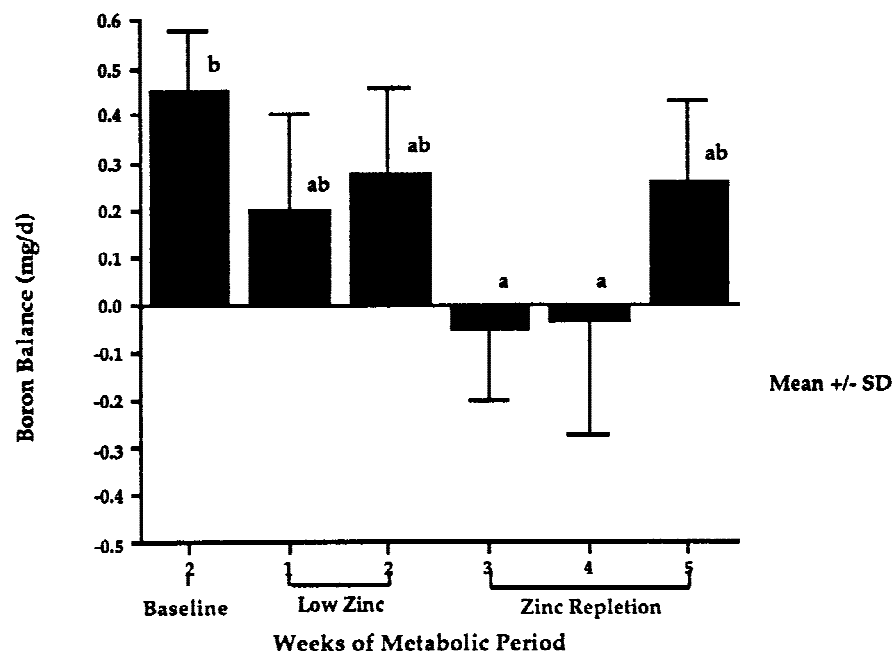


Fig. 5. Boron balance. (a), (b) and (ab) indicate significant statistical difference at  $P < 0.05$ .

ment, miscellaneous secretions, and blood draws would lower the net amount of boron retained overall.

### Urinary Calcium and Sodium

Urinary calcium excretion was higher during zinc repletion than during the second week of baseline and first 2 weeks of the low zinc period, 147 vs. 204 mg/d on average ( $P < 0.05$ ) (Table VI). Urinary sodium did not change during the course of the entire study; it averaged 2 g/d.

### DISCUSSION

The results of this study and that of Kent and McCance [3] and Hunt et al. [4] show that dietary boron is rapidly absorbed and excreted in the urine. A change in dietary boron from one day to the next caused a change in urinary boron excretion. However, the change in urinary boron was less than the change in boron intake; a 60% increase in dietary boron, 1.9–3.0 mg/d, caused only a 10% increase in urinary boron. Beattle and Peace [6] reported a similar response in postmenopausal women; when dietary boron was increased from 0.33 mg/d–3.33 mg/d, urinary boron excretion increased rapidly over the first 3 days after the dietary change and then stabilized at 90% of boron intake.

Urinary boron excretion tends to be 84–90% of boron intake irrespective of the amount of dietary boron consumed. When 3.23 mg of boron/d was fed to postmenopausal women [4], 89% was excreted in the urine and 3% in the feces. Naghii and Samman [7] reported that young men supplemented with 10 mg boron/d in addition

to their habitual diet excreted 84% of the total intake in the urine. Our subjects, consuming an average of 3.73 mg boron/d, excreted 85% of the intake in the urine and 8% in the feces. Urinary boron changes quickly with changes in dietary boron intake and, therefore, is a sensitive indicator of recent boron intake. We recently published a preliminary report of partial analyses of the present study [8].

Variation of daily urinary boron excretion (16–25%) in our subjects was similar to that seen for the daily urinary excretion of sodium (17–24%) and creatinine (7–18%). We consider this variation to be within the normal biological variation while on constant dietary intakes.

Our subjects were in a slight positive boron balance, an average of 0.19 mg boron/d, over a 6-week period consisting of the second week of baseline, the first 2 weeks of the low zinc period, and the last 3 weeks of the 5-week zinc repletion period. Hunt et al. [4] reported that when the dietary boron intake of postmenopausal women was 0.36 mg/d, the women were in a slight negative boron balance; an average of 0.06 mg boron was lost each day. When the dietary boron intake was increased to 3.23 mg/d, the women were in a slight positive boron balance with the retention of 0.25 mg boron/d. Neither of these studies considered the loss of boron via the integument, miscellaneous secretion and blood loss due to phlebotomy during the course of the study. There was a higher retention of boron during the first 2 weeks of the low zinc period, an average of 0.24 mg boron/d, than during the last 3 weeks of the zinc repletion period, when 0.06 mg boron/d was retained. The highest urinary boron loss occurred during the third and fourth weeks of zinc repletion, thus causing the decline in boron balance. The cause of this elevated level of boron excretion is not known. We found no evidence of analytical problems, and there was no association between the excretion of boron and zinc in the urine.

The average daily urinary calcium excretion was higher during the 5 weeks of zinc repletion than during the last week of the baseline period and the first 2 weeks of the low zinc period. Hunt et al. [4] reported a decrease in urinary calcium in postmenopausal women who were fed a low boron, low magnesium diet and supplemented with 3 mg boron/d. When the women were supplemented with magnesium and boron, urinary calcium excretion increased.

However, the effect of boron on bone mineral metabolism remains controversial. Beattie and Peace [6] reported no effect of boron on urinary calcium excretion in postmenopausal women when dietary boron intake was increased by 3 mg/d. Naghii and Samman [7] observed a decline in urinary calcium excretion in young men supplemented with 10 mg boron/d for 4 weeks. In our study, urinary calcium excretion was elevated during the zinc repletion period. No correlation was observed between urinary calcium and urinary boron losses. We do not believe that the increase in urinary calcium excretion seen in our study is associated with boron metabolism. Until the daily urinary calcium excretion values are available for the entire study, we cannot speculate on the cause of this increase in urinary calcium excretion.

## CONCLUSIONS

In summary, boron homeostasis appears to be primarily regulated by the kidney. Human studies indicate that urinary boron excretion reflects dietary intake and is a sensitive indicator of recent intakes over the range of 0.35–10.0 mg B/d. The vari-

ability in excretion of boron in the urine is similar to the that for sodium and creatinine in men on a constant diet. Urinary excretion of boron did not change with an increase or decrease in dietary zinc. Urinary boron excretion did increase during the third and fourth weeks of the 5-week zinc repletion period, but the cause of this increase is not known. Although urinary calcium excretion was higher during the entire 5 weeks of the zinc repletion period than earlier in the study, there was no association between urinary boron and calcium excretion.

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